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DB=USPT,PGPB,JPAB,EPAB,DWPI,TDBD; THES=ASSIGNEE; PLUR=YES; OP=AND			
L11	(allergen) same (hemagglutinin adj A)	1	L11
L10	L8 and (hemagglutinin adj A)	2	L10
L9	L8 and (hemagglutinin adj A)	2	L9
L8	L7 and (ragweed or pollen or (plant adj allergen))	116	L8
L7	L6 and L5	348	L7
L6	(heterologous or deletion or fusion) same L4	12311	L6
L5	L4 and (allergen)	753	L5
L4	(signal or leader) adj (sequence or peptide)	43160	L4
L3	L2 and ((signal or leader) adj (sequence or peptide))	2	L3
L2	L1 and (allergen)	14	L2
L1	Raz-Eyal.in.	28	L1

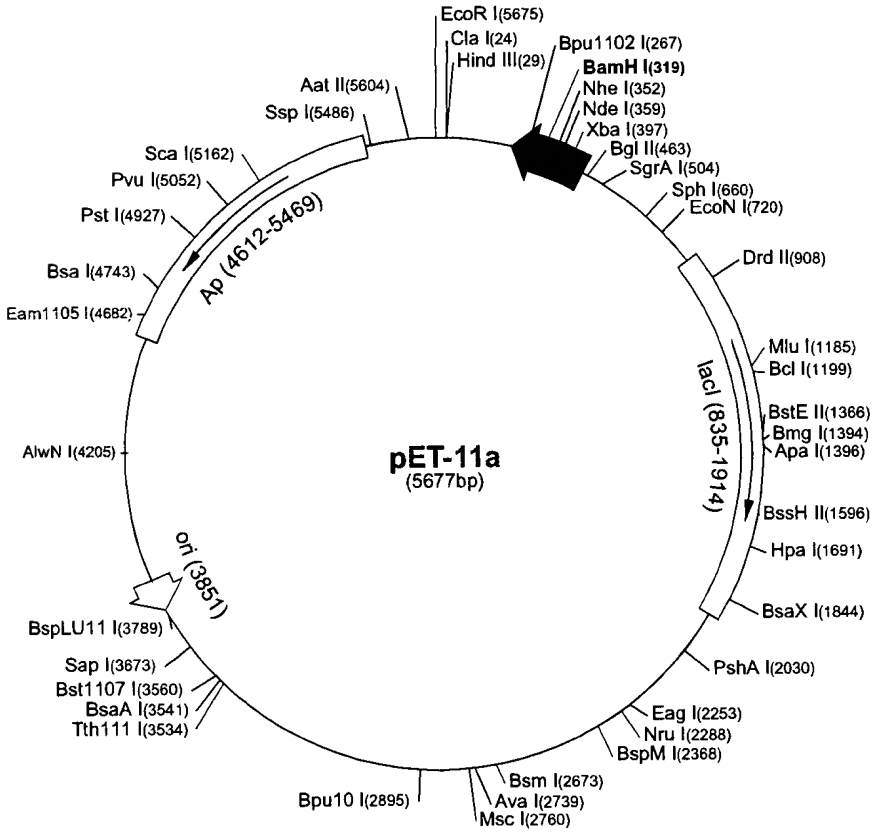
pET-11a-d Vectors

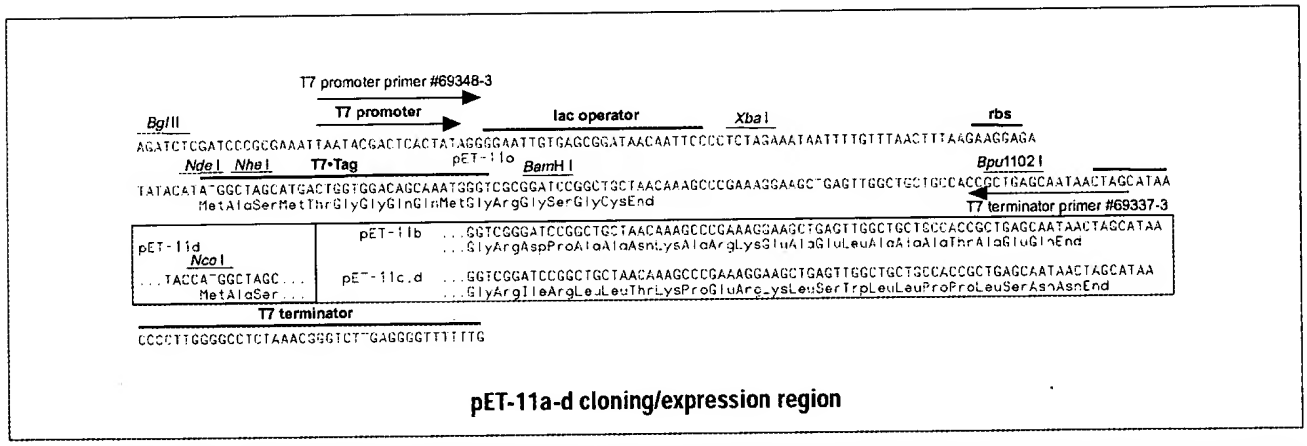
	Cat. No.
pET-11a DNA	69436-3
pET-11b DNA	69437-3
pET-11c DNA	69438-3
pET-11d DNA	69439-3

The pET-11a-d vectors carry an N-terminal T7•Tag® sequence and *Bam*H I cloning site. These vectors are the precursors to many pET family vectors; the pET-21a-d(+) series corresponds to pET-11a-d but incorporates several additional features. Unique sites are shown on the circle map. Note that the sequence is numbered by the pBR322 convention, so the T7 expression region is reversed on the circular map. The cloning/expression region of the coding strand transcribed by T7 RNA polymerase is shown below.

pET-11a sequence landmarks	
T7 promoter	432-448
T7 transcription start	431
T7•Tag coding sequence	328-360
T7 terminator	213-259
lacI coding sequence	835-1914
pBR322 origin	3851
bla coding sequence	4612-5469

The maps for pET-11b, pET-11c and pET-11d are the same as pET-11a (shown) with the following exceptions: pET-11b is a 5676bp plasmid; subtract 1bp from each site beyond *Bam*H I at 319. pET-11c is a 5675bp plasmid; subtract 2bp from each site beyond *Bam*H I at 319. pET-11d is a 5674bp plasmid; the *Bam*H I site is in the same reading frame as in pET-11c. An *Nco* I site is substituted for the *Nde* I site with a net 1bp deletion at position 359 of pET-11c. As a result, *Nco* I cuts pET-11d at 355. For the rest of the sites, subtract 3bp from each site beyond position 360 in pET-11a. *Nde* I does not cut pET-11d.





pET-11a-d cloning/expression region

pET-11a Restriction Sites

TB042 12/98

Enzyme	# Sites	Locations
AatII	1	5604
AccI	1	3559
AceIII	7	952 1680 2011 3298 3439 3741 4981
AciI	89	
AflIII	2	1185 3789
AluI	24	
AlwI	16	
Alw21I	8	685 1169 2492 2783 3607 4107 5268 5353
Alw44I	4	1165 3603 4103 5349
AlwNI	1	4205
ApaI	1	1396
ApaBI	2	869 2366
ApoI	2	1460 5675
AvaI	1	2739
AvaII	9	1737 2113 2201 2450 2753 2795 3074 4820 5042
BamHI	1	319
BanI	12	
BanII	3	569 583 1396
BbsI	5	1331 1670 2044 2907 5660
BbvI	27	
BccI	15	
Bce83I	7	208 1999 2169 3880 4178 4419 5287
BcefI	5	704 1045 1672 2481 4291
BcgI	10	315 349 1477 1511 2011 2045 3366 3400 5187 5221
BclI	1	1199
BfaI	7	257 353 398 2803 4284 4537 4872
BglI	3	2249 2483 4802
BglII	1	463
BmgI	1	1394
BpmI	6	1023 1512 2146 2700 3316 4752
Bpu10I	1	2895
Bpu1102I	1	267
BsaI	1	4743
BsaAI	1	3541
BsaBI	3	462 468 2986
BsaHI	8	508 529 643 1142 1825 2520 5219 5601
BsaJI	10	115 129 244 622 628 1820 2481 2683 2761 3949
BsaWI	7	189 1504 2007 2978 3995 4142 4973
BsaXI	1	1844
BsbI	2	3505 5225
BscGI	13	
BsgI	3	1036 1236 2949
BsiI	3	3962 5346 5653
BsiEI	6	1970 2256 3705 4129 5052 5201
BsII	22	
BsmI	1	2673
BsmAI	7	882 1287 1413 1800 3430 4743 5519
BsmBI	2	1800 3430
BsmFI	4	646 2187 2412 3060
BsoFI	50	
Bsp24I	14	
Bsp1286I	11	
BspEI	2	189 2978
BspGI	3	2373 2450 3315
BspLU11I	1	3789
BspMI	1	2368
BsrI	26	
BsrBI	3	418 3722 5523
BsrDI	4	1232 1598 4743 4917
BsrFI	8	160 495 504 871 2083 2243 2597 4762
BssHII	1	1596
Bst1107I	1	3560
BstEII	1	1366
BstXI	3	987 1116 1239
BstYI	11	
Cac8I	42	
CjeI	28	
CjePI	26	
ClaI	1	24
CviJI	95	
CviRI	26	
DdeI	11	
DpnI	29	
DraI	3	4548 4567 5259
DrdI	2	3482 3897
DrdII	1	908
DsaI	2	622 2761
EaeI	6	493 625 1859 2253 2758 5070
EagI	1	2253
Eam1105I	1	4682
EarI	3	803 3673 5477
EciI	5	962 2709 3863 4009 4837
Eco47III	3	590 2091 3043
Eco57I	2	4337 5349
EcoNI	1	720
EcoO109I	5	240 618 2753 2795 5658
EcoRI	1	5675
EcoRII	10	129 908 1223 1763 1820 2372 2755 3815 3936 3949
EcoRV	2	187 1635
FauI	18	
FokI	14	
FspI	3	2672 2770 4904
GdiII	5	493 625 1859 2253 5070
HaeI	8	913 2234 2306 2363 2760 3804 3815 4267
HaeII	13	
HaeIII	27	
HgaI	15	
HgiEII	2	783 4375
HhaI	44	
Hin4I	5	16 1084 2455 4681 4755
HincII	2	1691 5223
HindIII	1	29
HinfI	14	
HpaI	1	1691
HphI	17	
MaeII	12	
MaeIII	16	
MboII	15	
MluI	1	1185
MmeI	2	4004 4188
MnlI	33	
MscI	1	2760
MseI	24	
MslI	10	1237 1525 1555 2345 2776 2971 3362 4934 5093 5452
MspI	35	
MspA1I	10	271 1215 1785 1878 2455 3380 3499 4131 4376 5317
MwoI	45	
NarI	5	508 529 643 1825 2520
NciI	14	
NdeI	1	359
NgoAIV	4	495 2083 2243 2597
NheI	1	352
NlaIII	31	
NlaIV	28	
NruI	1	2288
NspI	4	660 3134 3426 3793
Pfl1108I	2	2072 4700
PflMI	3	767 2635 2684
PleI	7	446 734 821 1617 3683 4168 4671
PshAI	1	2030
Psp5II	2	2753 2795
Psp1406I	5	847 2215 3114 4908 5281
PstI	1	4927
PvuI	1	5052
PvuII	3	1785 1878 3380
RcaI	4	583 4509 5517 5622
RsaI	4	165 1332 3595 5162
SapI	1	3673
Sau96I	21	
Sau3AI	29	
ScaI	1	5162
ScrFI	24	
SfaNI	24	
SfcI	5	138 431 4054 4245 4923
SgrAI	1	504
SphI	1	660
SspI	1	5486
StyI	2	244 2683
TaqI	12	
TaqII	8	1093 1311 1984 3691 5030 5215 5368 5385
TfiI	7	1864 2166 2320 2618 2839 3343 3764
ThaI	40	
TseI	27	
Tsp45I	9	124 1366 2194 2461 3228 3441 3536 4938 5149
Tsp509I	16	
Tth111I	1	3534
Tth111II	7	1024 1717 3250 4379 4386 4418 5674
UbaJI	23	
VspI	4	446 1870 1929 4854
XbaI	1	397
XcmI	3	1041 1557 1575
XmnI	2	3347 5281

Enzymes that do not cut pET-11a:

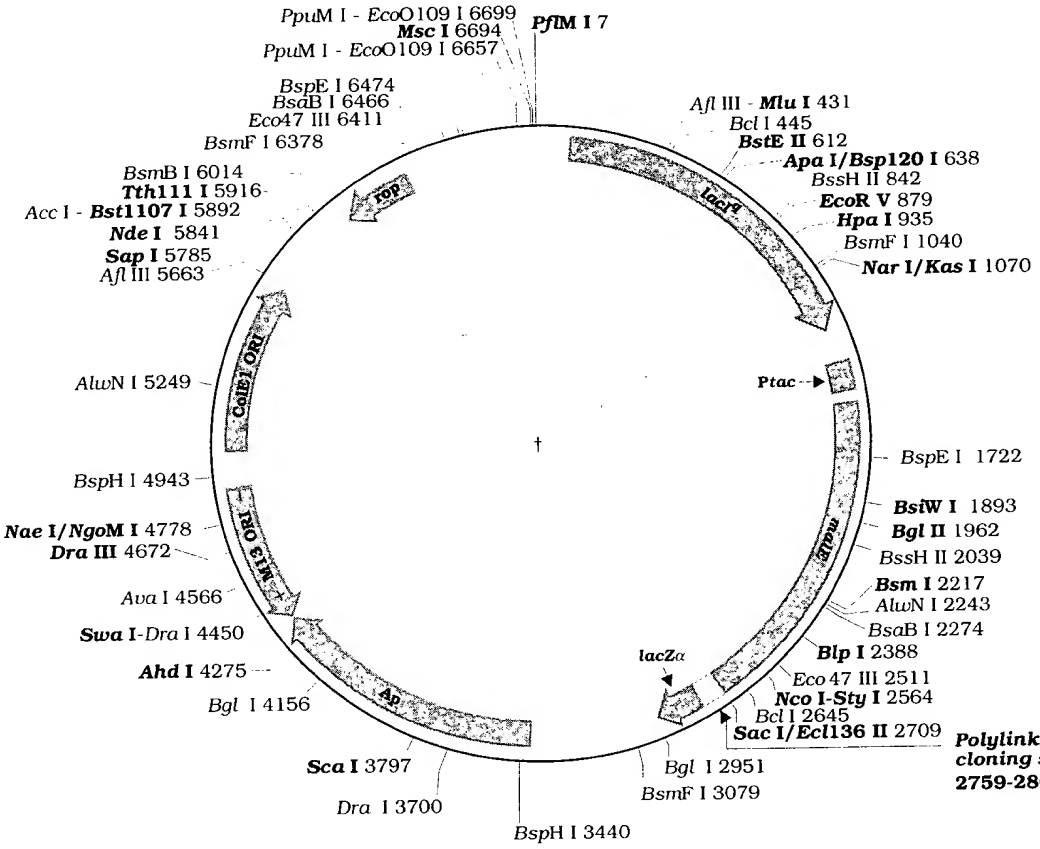
AflII	AgeI	AscI	AvrII	BaeI
BseRI	BsrGI	Bsu36I	DraIII	FseI
KpnI	MunI	NcoI	NotI	NsiI
NspV	PacI	PmeI	PmlI	RleAI
RsrII	SacI	SacII	SalI	SexAI
SfiI	SgfI	SmaI	SnaBI	SpeI
SrfI	Sse8387I	StuI	SunI	SwaI
XhoI				

pMAL™-p2

6,721 base pairs
See page 165 for ordering information.

pMAL™-p2 is an *E. coli* cloning vector used in the Protein Fusion and Purification System (#800, page 164). It is designed to create fusions between a cloned gene and the *E. coli malE* gene, which codes for maltose binding protein (MBP). The MBP fusion can then be expressed and purified, taking advantage of the properties of MBP. pMAL™-c2 is identical to pMAL™-p2 except for a deletion of the *malE* signal sequence (bases 1531–1605). The vectors contain the inducible P_{tac} promoter, positioned to transcribe a *malE-lacZα* gene fusion. The *lacI*^{q} gene encodes the Lac repressor, which turns off transcription from P_{tac} until IPTG is added. The polylinker provides restriction endonuclease sites to insert the gene of interest, fusing it to the *malE* gene. A portion of the *rrnB* operon containing two terminators, derived from the vector pKK233-2, prevents transcription originating from P_{tac} from interfering with plasmid functions. The gene for β-lactamase (Ap) and the origin of replication are from pBR322. The M13 origin is derived from pZ150. Nucleotide numbering starts at the beginning of the *lacI*^{q} fragment.

The map shows restriction sites of those enzymes that cut the molecule once or twice; the unique sites are shown in **bold** type. The table lists those sites that cut a moderate number of times. The polylinker is shown beneath the map. The coordinates refer to the position of the 5′ base in each recognition sequence. The map also shows the relative positions of the coding sequences and the origin of replication. The exact positions are: *lacI*^{q} 81–1161; P_{tac} 1406–1433; *malE* 1528–2703; polylinker 2704–2809; *lacZα* 2810–2968 (first in-frame stop codon 2989); β-lactamase (Ap) 3493-4351; M13 origin 4395-4908; ColE1 origin 5607.



Ecl136 II
Sac I
malE ... TCG AGC TCG AAC AAC AAC AAC AAT AAC AAT AAC AAC AAC CTC GGG

2760 2770 2780 2790 **Sse8387 I** 2800
Xmn I **EcoR I** **Xba I** **Pst I**
ATC GAG GGA AGG ATT TCA GAA TTC GGA TCC TCT AGA GTC GAC CTG CAG GCA AGC TTG .... lacZα
Ile Glu Gly Arg
BamH I **Sal I** **Hind III**

Factor Xa cleavage site

References

1. Guan, C., Li, P., Riggs, P.D. and Inouye, H. (1987) *Gene* 67, 21–30.
2. Maina, C.V. et al (1988) *Gene* 74, 365–373.
3. Riggs, P., in Ausubel, F.M. et al. (eds), *Current Prot. in Molecular Biol.* (1992) Greene Associates/Wiley Interscience, New York.
4. Zagursky, R.J. and Berman, M.L. (1984) *Gene* 27, 183–191.

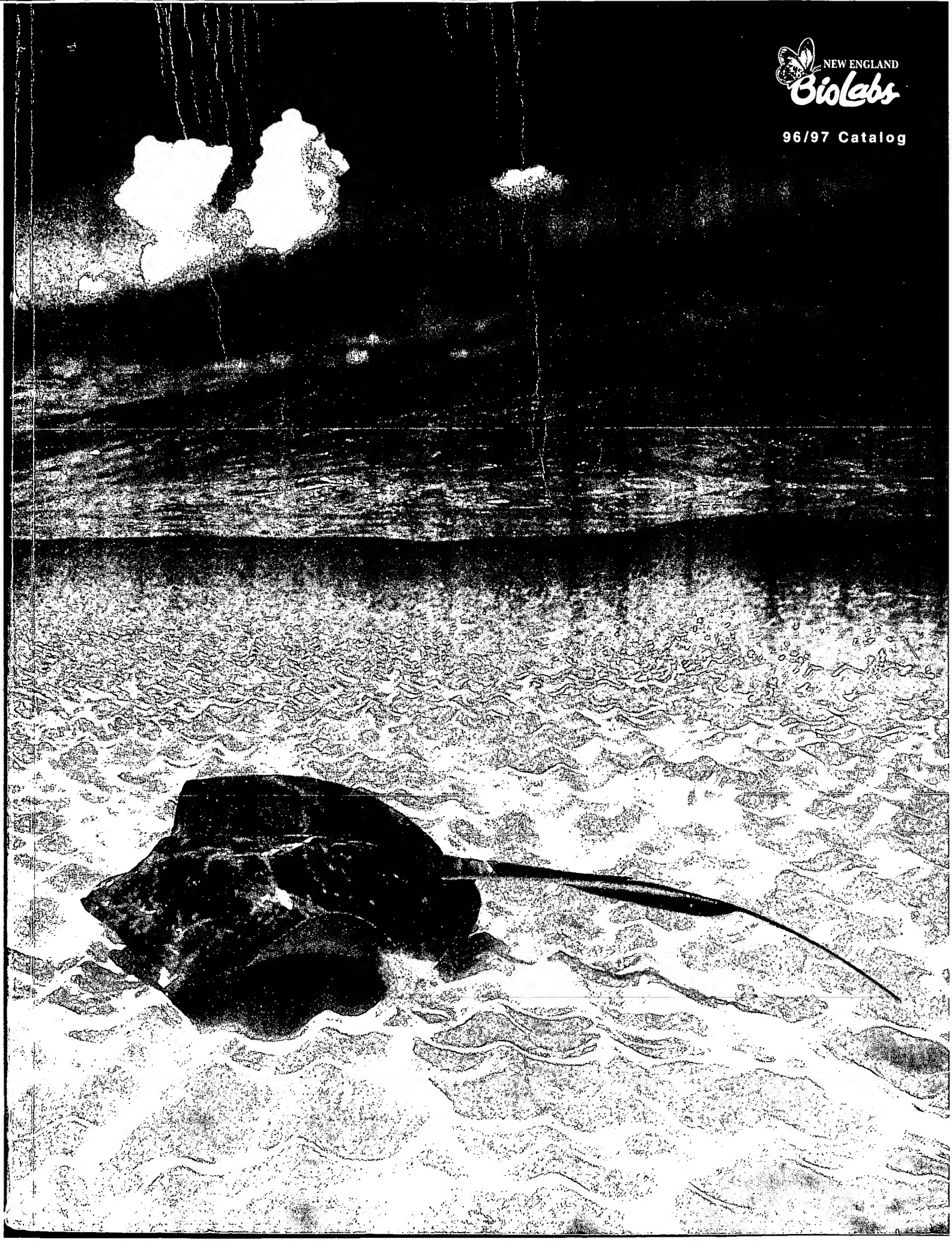
pMAL™-p2 DNA: Location of Sites

Enzyme	#	Locations			
Ahd I/*Eam*1105 I	1	4275			
Apa I/*Bsp*120 I	1	638			
*Bam*H I	1	2776			
Bgl II	1	1962			
Blp I	1	2388			
*Bsi*W I	1	1893			
Bsm I	1	2217			
*Bsp*LU11 I	1	5663			
*Bst*1107 I	1	5892			
*Bst*E II	1	612			
Dra III	1	4672			
*Ecl*136 II/*Sac* I	1	2709			
*Eco*R I	1	2770			
*Eco*R V	1	879			
Hind III	1	2802			
Hpa I	1	935			
Kas I/*Nar* I	1	1070			
Mlu I	1	431			
Msc I	1	6694			
Nae I/*Ngo*M I	1	4778			
Nco I	1	2564			
Nde I	1	5841			
*Pfl*M I	1	7			
Pst I	1	2794			
Sal I	1	2788			
Sap I	1	5785			
Sca I	1	3797			
*Sse*8387 I	1	2793			
Sty I	1	2564			
Swa I	1	4449			
*Tth*111 I	1	5916			
Xba I	1	2782			
Xmn I	1	2759			
Acc I	2	2788	5892		
Afl III	2	431	5663		
*Alw*N I	2	2243	5249		
Ava I/*Bso*B I	2	2746	4566		
Bcl I	2	445	2645		
Bgl I	2	2951	4156		
*Bsa*B I	2	2274	6466		
*Bsm*B I	2	1040	6014		
*Bsm*F I	2	3079	6378		
*Bsp*E I	2	1722	6474		
*Bsp*H I	2	3440	4943		
*Bss*H II	2	842	2039		
Dra I	2	3700	4450		
*Eco*47 III	2	2511	6411		
*Eco*O109 I	2	6657	6699		
*Ppu*M I	2	6657	6699		
Ban II	3	638	2709	4748	
Bbs I	3	570	909	6538	
Bcg I	3	734	3753	6067	
*Bsa*A I	3	2549	4675	5911	
Bsa I	3	1660	3140	4208	
*Bsr*F I	3	117	4195	4778	
*Bss*S I	3	1496	3611	5490	
Drd I	3	4626	5555	5968	
Dsa I	3	2290	2564	6691	
Fsp I	3	2945	4055	6684	
Nsp I	3	5663	6028	6322	
Pvu I	3	1925	2925	3908	
Xcm I	3	280	796	814	

Enzyme	#	Locations				
Ase I	4	1115	1174	1412	4104	
Bpm I	4	284	737	4190	6155	
*Bsp*M I	4	1294	2054	2395	2792	
*Bsr*D I	4	479	837	4039	4221	
*Bst*X I	4	226	355	478	1981	
*Eco*57 I	4	1667	2515	3593	5136	
Pvu II	4	1029	1122	2895	6072	
Ssp I	4	1387	3473	4446	4467	
Tfi I	4	1110	5689	6108	6614	
Ban I	5	351	1070	2503	4327	4714
*Bsa*H I	5	387	1070	1257	3318	3739
*Bsr*B I	5	1446	3270	3436	4819	5732
Eae I	5	1105	2077	2813	3887	6694
Sfc I	5	2794	4034	4896	5207	5398
*Apa*L I	6	411 5847	1241	1544	3608	5349
Bfa I	6	2783 6651	4087	4828	4917	5170
Bsg I	6	297 6488	497	1234	1292	6101
Hinc II	6	935 3736	1405	2191	2360	2788
*Psp*1406 I	6	92 6339	3281	3677	4050	4461
*Bsa*W I	7	750 5457	1696 6474	1722	3984	5310
Ear I	7	54 3486	1735 5785	1993	2530	2911
*Bsi*E I	8	1797 3908	1925 5326	2661 5750	2925	3759
*Csp*6 I/*Rsa* I	8	577 2636	1894 3798	2067 5858	2130	2341
Ple I	8	59 4626	855 5293	2786 5764	4281	4604
*Alw*26 I	9	123 3141	528 3444	654 4209	1041 6015	1661
Apo I	9	706 4479	1681 4490	1741 6248	2770 6717	3051
Ava II	9	983 4138	1516 6379	2300 6658	3078 6700	3916
*Tsp*45 I	9	613 4851	1708 5914	2834 6009	3808 6222	4019
*Bsi*HKA I	10	411 3608	1241 3693	1499 5349	1544 5847	2709 6673
Msl I	10	479 3864	767 4023	797 6088	2212 6481	3505 6676
*Bst*Y I	11	1962 3650 6471	1997 4913	2544 4925	2776 5011	3633 5022
*Bsa*J I	12	1066 2290 5503	1592 2564 6691	1844 2745	1883 2847	1988 4396
Dde I	12	1003 3779 6396	2389 4319 6558	3102 4980	3192 5389	3248 5854

There are no restriction recognition sites for the following enzymes:

Aat II, *Acc*65 I/*Kpn* I, *Afl* II, *Age* I, *Asc* I, *Avr* II, *Bse*R I, *Bsp*D I/*Cla* I, *Bsr*G I, *Bst*B I, *Bsu*36 I, *Eag* I, *Eco*N I, *Fse* I, *Mfe* I, *Nhe* I, *Not* I, *Nru* I, *Nsi* I/*Ppu*10 I, *Pac* I, *Pae*R7 I/*Xho* I, *Pme* I, *Pml* I, *Psh*A I, *Rsr* II, *Sac* II, *Sex*A I, *Sfi* I, *Sgf* I, *Sgr*A I, *Sma* I/*Xma* I, *Sna*B I, *Spe* I, *Sph* I, *Srf* I, *Stu* I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pGEX Vectors (GST Gene fusion)

ORDERING INFORMATION

Product	Quantity	Code Number
Glutathione S-transferase Gene Fusion Vectors*		
pGEX-1λT *Eco*R I/BAP	5 µg	27-4805-01
pGEX-2T	25 µg	27-4801-01
pGEX-2TK	25 µg	27-4587-01
pGEX-3X	25 µg	27-4803-01
pGEX-4T-1	25 µg	27-4580-01
pGEX-4T-2	25 µg	27-4581-01
pGEX-4T-3	25 µg	27-4583-01
pGEX-5X-1	25 µg	27-4584-01
pGEX-5X-2	25 µg	27-4585-01
pGEX-5X-3	25 µg	27-4586-01
pGEX-6P-1	25 µg	27-4597-01
pGEX-6P-2	25 µg	27-4598-01
pGEX-6P-3	25 µg	27-4599-01

***** *All vectors include E. coli BL21 cells.***

All of the GST gene fusion vectors offer:

- ***A tac promoter for chemically inducible, high-level expression.***
- **An internal *lac* I^q gene for use in any *E.* host.**
- **Very mild elution conditions for release proteins from the affinity matrix, thus**

minimizing effects on antigenicity and fu activity.

- PreScission™, thrombin, or factor Xa prc recognition sites for cleaving the desire from the fusion product.

Thirteen pGEX vectors are available (see fig Nine of the vectors have an expanded multi cloning site (MCS) that contains six restricti The expanded MCS facilitates the unidirectic cloning of cDNA inserts obtained from librar constructed using many available lambda ve including λ ExCell Cloning Vector (27-5013-(27-5011-01; see λ ExCell *Not* I/*Eco*R I/CIP λ ExCell *Eco*R I/CIP for more details) and La ZAP. pGEX-6P-1, pGEX-6P-2, and pGEX-6P-3 encode the recognition sequence for site-sp cleavage by PreScission™ Protease; see PreScission™ Protease) between the GST dc and the multiple cloning site. pGEX -4T-1, pGEX-4T-2, and pGEX-4T-3 are derived from pGEX-2T and contain a thrombin recognitior pGEX-5X-1, pGEX-5X-2, and pGEX5X-3 are derivatives of pGEX-3X and possess a factor recognition site.

pGEX-2TK is uniquely designed to allow the detection of expressed proteins by directly l the fusion products *in vitro* (1). This vector the recognition sequence for the catalytic su cAMP-dependent protein kinase obtained fr muscle. The protein kinase site is located be the GST domain and the MCS. Expressed pro can be directly labelled using protein kinase [γ-^{32}P]ATP and readily detected using stan radiometric or autoradiographic techniques. pGEX-2TK is a derivative of pGEX-2T; its fus proteins can be cleaved with thrombin.

Cleavage of pGEX-6P GST fusion proteins oc

between the Gln and Gly residues of the rec

sequence Leu-Glu-Val-Leu-Phe-Gln-Gly-Pro temperature (5°C) digestion minimizes the degradation of the protein of interest. Becau PreScission™ Protease has been engineered GST-tag, it can also be removed from the cle mixture simultaneously with the GST portio fusion protein. The pGEX-6P Expression Vec permit convenient site-specific cleavage and simultaneous purification on Glutathione Sepharose™. The pGEX-6P series provides a translational reading frames linked betweer GST coding region and the multiple cloning

Collectively, the pGEX vectors provide all th translational reading frames beginning with EcoR I restriction site. pGEX-1λT, pGEX-6P-1 pGEX-4T-1, and pGEX-5X-1 can directly acce express cDNA inserts isolated from λ gt11 li

Click on "ASCII" to download an unformatte sequence for use by a sequence analysis pro Click on "PDF" to download a formatted seq and restriction site table. If you prefer acce sequence in ***GenBank***, refer to the right-han column for the GenBank accession number:

			GenBa
Vector	**Unformatted**	**Formatted**	**Acces**
pGEX-4T-1, 27-4580-01	*ASCII*	*PDF*	U1385
pGEX-4T-2, 27-4581-01	*ASCII*	*PDF*	U1385
pGEX-4T-3, 27-4583-01	*ASCII*	*PDF*	U1385
pGEX-5X-1, 27-4584-01	*ASCII*	*PDF*	U1385
pGEX-5X-2, 27-4585-01	*ASCII*	*PDF*	U1385
pGEX-5X-3, 27-4586-01	*ASCII*	*PDF*	U1385
pGEX-2TK, 27-4587-01	*ASCII*	*PDF*	U1385
pGEX-2T, 27-4801-01	*ASCII*	*PDF*	U1385
pGEX-3X, 27-4803-01	*ASCII*	*PDF*	U1385
pGEX-1 lambda T, 27-4805-01	*ASCII*	*PDF*	U1384
pGEX-6P-1, 27-4597-01	*ASCII*	*PDF*	U7887
pGEX-6P-2, 27-4598-01	*ASCII*	*PDF*	U7887
pGEX-6P-3, 27-4599-01	*ASCII*	*PDF*	U7887

Properties of pGEX Vectors . Induction: tac p inducible with 1-5 mM IPTG.

- **Expression: Proteins are expressed as fu proteins with the 26 kDa glutathione S-transferase (GST). The GST gene cont ATG and ribosome-binding site, and is u control of the tac promoter. A translatio terminator is provided in each reading fr The resulting fusion protein may be puri using the GST Purification Module (27-4 -02; see GST Purification Modules.)**
- **Enzymatic cleavage with PreScission™ P pGEX-6P-1, -2, -3 allow for removal of th carrier protein from the fusion protein b enzymatic cleavage with PreScission™ P Because PreScission™ Protease has bee engineered with a GST-tag, it can also b removed simultaneously with the GST pr the fusion protein.**
- **Enzymatic cleavage with thrombin: pGE**

lambda T, pGEX-2T, pGEX-2TK, pGEX-4T
-3 allow for removal of the GST carrier p
from the fusion protein by enzymatic cle
with thrombin.

- Enzymatic cleavage with factor Xa: pGE)
pGEX-5X-1, -2, -3 allow for removal of t
carrier protein from the fusion protein b
enzymatic cleavage with factor Xa.
- Direct labelling *in vitro*: pGEX-2TK allow
direct labelling of fusion proteins in vitr
32P using the catalytic subunit of
cAMP-dependent protein kinase.
- Host(s): *E. coli*. The plasmid provides la
repressor.
- Selectable marker(s): Plasmid confers
resistance to 100 µg/ml ampicillin.
- Amplification: Recommended.

. pGEX-2T Control Regions:

* Glutathione S-transferase gene region: *t*
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region fo
thrombin cleavage: 918-935

* MCS: 930-945

* Beta-lactamase gene region: Promoter: -
1309-1314; -35: 1286-1291; Start codon (A
1356; Stop codon (TAA): 2214

* *lac*Iq gene region: Start codon (GTG): 32
codon (TGA): 4377

* Plasmid replication region: Site of replic
initiation: 2974; Region necessary for replic
2281-2977

* Sequencing primers: 5' pGEX Sequencing
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1020-998

. pGEX-2TK Control Regions:

* Glutathione S-transferase gene region: *t*
promoter: -10: 205-211; -35: 183-188; *lac* o

217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region fo
thrombin cleavage: 918-935;
*** Coding for kinase recognition site: 936-9**
*** MCS: 951-966**
*** Beta-lactamase gene region: Promoter: -**
1330-1335; -35: 1307-1312; Start codon (A
1377; Stop codon (TAA): 2235
*** *lac*Iq gene region: Start codon (GTG): 33**
codon (TGA): 4398
*** Plasmid replication region: Site of replic**
initiation: 2995; Region necessary for replic
2302-2998
Sequencing primers: 5' pGEX Sequencing Pr
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1041-1019

• pGEX-3X Control Regions:
*** Glutathione S-transferase gene region: *t***
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region fo
Xa cleavage: 921-932
*** MCS: 934-949**
*** Beta-lactamase gene region: Promoter: -**
1313-1318; -35: 1290-1295; Start codon (A
1360; Stop codon (TAA): 2218
*** *lac*Iq gene region: Start codon (GTG): 33**
codon (TGA): 4381
*** Plasmid replication region: Site of replic**
initiation: 2978; Region necessary for replic
2285-2981
*** Sequencing primers: 5' pGEX Sequencing**
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1024-1002

• pGEX-1 Lambda T Control Regions:
*** Glutathione S-transferase gene region: *t***
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24

codon (ATG) for GST: 258; Coding region fo
thrombin cleavage: 918-935
* MCS: 930-944
* Beta-lactamase gene region: Promoter: -
1308-1313; -35: 1285-1290; Start codon (A
1355; Stop codon (TAA): 2213
* *lac*Iq gene region: Start codon (GTG): 32
codon (TGA): 4376
* Plasmid replication region: Site of replic
initiation: 2973; Region necessary for replic
2280-2976
* Sequencing primers: 5' pGEX Sequencing
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1019-997

• pGEX-4T-1 Control Regions:
* Glutathione S-transferase gene region: *t*
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region fo
thrombin cleavage: 918-935
* MCS: 930-966
* Beta-lactamase gene region: Promoter: -
1330-1335; -35: 1307-1312; Start codon (A
1377; Stop codon (TAA): 2235
* *lac*Iq gene region: Start codon (GTG): 33
codon (TGA): 4398
* Plasmid replication region: Site of replic
initiation: 2995; Region necessary for replic
2302-2998
* Sequencing primers: 5' pGEX Sequencing
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1041-1019

• pGEX-4T-2 Control Regions:
* Glutathione S-transferase gene region: *t*
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region fo
thrombin cleavage: 918-935

* MCS: 930-967
* Beta-lactamase gene region: Promoter: -1331-1336; -35: 1308-1313; Start codon (A 1378; Stop codon (TAA): 2236
* *lac*Iq gene region: Start codon (GTG): 33 codon (TGA): 4399
* Plasmid replication region: Site of replica initiation: 2996; Region necessary for replic 2303-2999
* Sequencing primers: 5' pGEX Sequencing binds nucleotides 869-891; 3' pGEX Sequen Primer binds nucleotides 1042-1020

• pGEX-4T-3 Control Regions:
* Glutathione S-transferase gene region: *t* promoter: -10: 205-211; -35: 183-188; *lac* o 217-237; Ribosome binding site for GST: 24 codon (ATG) for GST: 258; Coding region fo thrombin cleavage: 918-935
* MCS: 930-965
* Beta-lactamase gene region: Promoter: -1329-1334; -35: 1306-1311; Start codon (A 1376; Stop codon (TAA): 2234
* *lac*Iq gene region: Start codon (GTG): 33 codon (TGA): 4397
* Plasmid replication region: Site of replica initiation: 2994; Region necessary for replic 2301-2997
* Sequencing primers: 5' pGEX Sequencing binds nucleotides 869-891; 3' pGEX Sequen Primer binds nucleotides 1040-1018

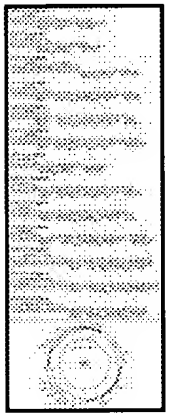
• pGEX-5X-1 Control Regions:
* Glutathione S-transferase gene region: *t* promoter: -10: 205-211; -35: 183-188; *lac* o 217-237; Ribosome binding site for GST: 24 codon (ATG) for GST: 258; Coding region fo Xa cleavage: 921-932
* MCS: 934-969
* Beta-lactamase gene region: Promoter: -

1333-1338; -35: 1310-1315; Start codon (A
1380; Stop codon (TAA): 2238
*** *lac*Iq gene region: Start codon (GTG): 33**
codon (TGA): 4401
*** Plasmid replication region: Site of replica**
initiation: 2998; Region necessary for replic
2305-3001
*** Sequencing primers: 5' pGEX Sequencing**
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1044-1022

• pGEX-5X-2 Control Regions:
*** Glutathione S-transferase gene region: *ta***
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region for
Xa cleavage: 921-932
*** MCS: 934-970**
*** Beta-lactamase gene region: Promoter: -**
1334-1339; -35: 1311-1316; Start codon (A
1381; Stop codon (TAA): 2239
*** *lac*Iq gene region: Start codon (GTG): 33**
codon (TGA): 4402
*** Plasmid replication region: Site of replica**
initiation: 2999; Region necessary for replic
2306-3002
*** Sequencing primers: 5' pGEX Sequencing**
binds nucleotides 869-891; 3' pGEX Sequen
Primer binds nucleotides 1045-1023

• pGEX-5X-3 Control Regions:
*** Glutathione S-transferase gene region: *ta***
promoter: -10: 205-211; -35: 183-188; *lac* o
217-237; Ribosome binding site for GST: 24
codon (ATG) for GST: 258; Coding region for
Xa cleavage: 921-932
*** MCS: 934-971**
*** Beta-lactamase gene region: Promoter: -**
1335-1340; -35: 1312-1317; Start codon (A
1382; Stop codon (TAA): 2240

* ***lacI*q gene region: Start codon (GTG): 33 codon (TGA): 4403**
* **Plasmid replication region: Site of replica initiation: 3000; Region necessary for replic 2307-3003**
* **Sequencing primers: 5' pGEX Sequencing binds nucleotides 869-891; 3' pGEX Sequen Primer binds nucleotides 1046-1024**



Click here for *PDF version of image*

Map of the glutathione S-transferase fusion showing the reading frames and main featu Even though stop codons in all three frames depicted in this map, all thirteen vectors ha codons in all three frames downstream from multiple cloning site.

References

1. Kaelin, W.G. *et al., Cell 70,* 351 (1992).

Related Products	Code Number
GST Purification Modules	
GST Detection Module (50 detections)	27-4590-01
GSTrap™ FF Columns	
GSTPrep FF 16/10	17-5234-01
Glutathione Sepharose™ 4B Lab Packs and Columns	
PreScission Protease	27-0843-01
Anti-GST Antibody (50 detections)	27-4577-01
GST Vector Primers for Sequencing	
E. coli BL21	27-1542-01